

WHAT IS CLAIMED IS:

1. An isolated nucleic acid encoding an antifreeze protein, said protein defined as follows:
 - (i) having a calculated molecular weight of between 7 and 13 kDa;
 - (ii) having a thermal hysteresis activity greater than about 1 °C at about 1 mg/mL; and
 - (iii) (a) specifically binding to an antibody raised against antifreeze proteins selected from the group consisting of YL-1, YL-2, YL-3 or YL-4; or (b) having at least 60% amino acid sequence identity to antifreeze proteins selected from the group consisting of YL-1, YL-2 YL-3 or YL-4; or (c) having an amino acid sequence comprising at least one repeating unit of the consensus sequence of SEQ ID NO: 1
2. The isolated nucleic acid of claim 1, wherein the calculated molecular weight of the encoded protein is between about 8 and 12 kDa.
3. The isolated nucleic acid of claim 1, wherein the thermal hysteresis activity is greater than 2°C at 1 mg/mL.
4. The isolated nucleic acid of claim 1, wherein the encoded protein has at least 80% sequence identity to antifreeze proteins selected from the group consisting of YL-1, YL-2, YL-3 and YL-4.
5. The isolated nucleic acid of claim 1, wherein the encoded protein is selected from the group consisting of YL-1, YL-2, YL-3, and YL-4.
6. The isolated nucleic acid of claim 1, wherein the encoded protein consists of about 5 to 12 repeating units of at least 5 out of 6 conserved amino acids of SEQ ID NO: 1.

7 An isolated nucleic acid which specifically hybridizes to SEQ ID NO:2 under stringent conditions

8 An isolated nucleic acid encoding at least 1 repeat of a 12 contiguous amino acid motif from a purified *Tenebrio molitor* antifreeze protein which specifically binds to an antibody directed against antifreeze proteins selected from the group consisting of YL-1, YL-2 YL-3 and YL-4

9. An isolated antifreeze protein, said protein:

- (i) having at least 1 repeat of a 12 contiguous amino acid motif comprising SEQ ID NO:1;
- (ii) having a calculated molecular weight of about 7.0 to 13.0 kDa;
- (iii) having a pI of about 8.0 to 10.0,
- (iv) having a thermal hysteresis activity of greater than about 1.5°C at about 1 mg/mL; and
- (v)
 - (a) having the amino acid motif as shown in SEQ ID NO:3 at the N-terminus; or
 - (b) specifically binding to antibodies raised to antifreeze proteins selected from the group consisting of YL-1, YL-2, YL-3 and YL-4, or
 - (c) having 60% amino acid sequence identity to antifreeze proteins selected from the group consisting of YL-1, YL-2, YL-3 and YL-4

10. The isolated antifreeze protein of claim 9, wherein the number of repeating motifs is from 5 to 12

11. The isolated antifreeze protein of claim 9, wherein the thermal hysteresis activity is greater than about 2.0°C at a concentration of about 1 mg/mL.

12. The isolated antifreeze protein of claim 9, wherein the protein is selected from the group consisting of YL-1, YL-2, YL-3 and YL-4

13. An antibody, specifically immunoreactive under immunologically reactive conditions, to an antifreeze protein, said protein having at least 1 repeat of a sequence comprising the motif of SEQ ID NO:1

14. The antibody of claim 13, wherein the antifreeze protein further comprises the amino acid sequence SEQ ID NO:4.

15. An organism into which, or into an ancestor of which, an exogenous nucleic acid sequence which specifically hybridizes under stringent conditions to SEQ ID NO:4 has been introduced and the organism translates the exogenous nucleic acid into an antifreeze protein.

16. The organism of claim 15, wherein the exogenous nucleic acid sequence is translated into an antifreeze protein which is expressed externally from the organism.

17. The organism of claim 15, wherein said organism is a fish.

18. The organism of claim 18, wherein said fish is a member of the family Salmonidae.

19. The organism of claim 15, wherein said organism is a crustacean.

20. The organism of claim 19, wherein said organism is a member of the suborder Natantia.

21. The organism of claim 20, wherein said organism is selected from the group consisting of the genera *Palaemonetes* and *Penaeus*.

22. The organism of claim 19, wherein said organism is a member of the genus *Homarus*.

- 1 23. The organism of claim 15, wherein said organism is a plant
- 1 24. The organism of claim 15, wherein said organism is a fungus.
- 1 25. The organism of claim 24, wherein the fungus is a yeast.
- 1 26. The organism of claim 25, wherein the yeast is selected from the
2 group consisting of *Torulopsis holmii*, *Saccharomyces fragilis*, *Saccharomyces cerevisiae*,
3 *Saccharomyces lactis*, and *Candida pseudotropicalis*.
27. The organism of claim 15, wherein the organism is a bacteria.
28. The organism of claim 27, wherein the bacteria is selected from the
group consisting of *Streptococcus cremoris*, *Streptococcus lactis*, *Streptococcus*
thermophilus, *Leuconostoc citrovorum*, *Leuconostoc mesenteroides*, *Lactobacillus*
acidophilus, *Lactobacillus lactis*, *Bifidobacterium bifidum*, *Bifidobacterium breve*, and
Bifidobacterium longum.
29. A method for decreasing the freezing point of an aqueous solution,
2 said method comprising the addition of an antifreeze protein having greater than 1 repeat of
3 the contiguous amino acid motif of SEQ ID NO.1, to said aqueous solution.
- 1 30. The method of claim 29, wherein the aqueous solution is applied to
2 an organism
- 1 31. The method of claim 29, wherein the number of repeating motifs is
2 between about 5 to about 12.
- 1 32. The method of claim 29, wherein the antifreeze protein is produced
2 by recombinant means.

1 33 The method of claim 29, wherein the antifreeze protein further
2 specifically binds to an antibody directed against YL-1, YL-2, YL-3 or YL-4

1 34. The method of claim 29, wherein the antifreeze protein is selected
2 from the group consisting of YL-1, YL-2, YL-3, and YL-4.

1 35. The method of claim 32, wherein the antifreeze protein is encoded by
2 a nucleic acid molecule which specifically hybridizes to SEQ ID NO 2

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